UNIVERSITI PUTRA MALAYSIA

ENHANCEMENT OF PHOSPHORUS SOLUBILIZATION FROM PHOSPHATE ROCKS AND PLANT NUTRIENT AVAILABILITY THROUGH VERMICOMPOSTING

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By

TENGKU SABRINA DJUNITA

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirements for the Degree of Doctor of Philosophy

April 2007

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Doctor of Philosophy

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Chairman : Professor Mohamed Hanafi Musa, PhD, *AMIC*

Faculty : Agriculture

Large amount of organic materials (OM), from oil palm by-products produced by plantations in Malaysia creates potential pollutants and habitat for certain parasitic insects and pathogen. Incorporation of these materials with phosphate rock (PR) and earthworm through vermicomposting process would promote the dissolution and plant availability of phosphorus (P) from PR. This study investigated the effect of oil palm by-products and earthworms in dissolution different types of PRs for production of vermiphosphocompost (VPC) in fulfilling the P requirement of *Setaria splendida* in comparison to the using of arbuscular mycorrhiza (AM) in a glasshouse. The potential of earthworm in using oil palm by-products was investigated by surveying the population and diversity of earthworms in oil palm plantation with different types of soils and palm tree ages. Only exotic endogeic *Pontoscolex corethrurus* was found in oil palm plantation with low population (0 - 42 individual m^{-2}). Heterogeneity of earthworm population in oil palm plantation attributed to food and soil physical habitat as determined by a principal component analysis (PCA). Vermicomposting of PR mixed EFB using *Eisenia fetida* in the laboratory

gave a higher dissolution of P with morocco PR (MPR) >gafsa PR (GPR) > togo PR (TPR) than that of normal compost system. The corresponding values for frond were 165, 52, and 30%. This was attributed to higher humic acid content in VPC (0.07 g), the population of bacteria, extractable Ca, and enzymes phosphomonoesterase in the gut of earthworm. The bacteria in EFB-earthworm intestine was identified as *Pseudomonas nitroreducens,* and *P. citronellolis,* and *Cellulomonas flavigena* in frond-reared earthworm. Fresh EFB contain a significant value of total extractable phenol (10 g GAE 100 g^{-1} extract) at the beginning of composting process and decreased gradually after 4 weeks decomposition. The compound identified in fresh EFB was 2,4-bis(1,1-dimethyl)phenol, composted and field composted EFB was 2,6-bis(1,1-dimethyl) phenol. In contrast, there was no phenol detected in vermicomposted EFB. The direct application of 3 types earthworms in the glasshouse study using *Setaria* grass did not gave a significant difference on P availability, total nutrients in soil, nutrient-uptake and dry matter (DM) yield of grass. The VPC showed better growth of *Setaria* grass compared to EFB and conventional compost. *Setaria* needed about 75 ton ha^{-1} VPC to achieve a maximum DM for this experiment Inorganic fertilizer (GPR) mixed with EFB, AM and worm enhanced *Setaria* grass yield (60%) compare to GPR only and by using VPC (79%). The contribution of AM, earthworm of GPR on P uptake was varied depend on the interaction among them. In conclusion, VPC is the best way to managing EFB. Its nutrients content can support *Setaria* growth. Improvement of *Setaria* grass growth and plant availability of P and other nutrients (N and K) as indicated by nutrients uptake was obtained by mixing EFB with worm, AM, and inorganic fertilizers.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

PENINGKATAN PELARUTAN FOSFORUS DARI BATUAN FOSFAT DAN KETERSEDIAAN NUTRIEN BAGI TANAMAN MELALUI VERMIKOMPOS

Oleh

TENGKU SABRINA DJUNITA

April 2007

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Jumlah bahan organik (OM) yang banyak dari produk sampingan kelapa sawit yang dikeluarkan oleh ladang di Malaysia berpotensi menjadi bahan cemar dan habitat bagi serangga parasitik dan patogen tertentu. Pencampuran bahan-bahan tersebut dengan batuan fosfat (PR) dan cacing tanah melalui proses vermikompos akan meningkatkan pelarutan dan fosforus tersedia (P) bagi tanaman dari PR. Kajian ini meneneliti pengaruh hasil sampingan kelapa sawit dan cacing tanah untuk melarutkan berbagai jenis PRs untuk menghasilkan vermiphosphokompos (VPC) bagi memenuhi keperluan P untuk *Setaria splendida* dibandingkan dengan penggunaan arbuskular mikoriza (AM) di rumah kaca. Potensi menggunakan cacing tanah pada hasil sampingan kelapa sawit diuji dengan tinjauan populasi dan kepelbagaian cacing tanah di ladang kelapa sawit pada berberapa jenis tanah dan umur pokok kelapa sawit yang berbeza. Hanya cacing endogeic eksotik *Pontoscolex corethrurus* ditemui di ladang kelapa sawit dengan populasi yang rendah (0 – 42 individu m^{-2}). Kepelbagaian populasi cacing tanah di ladang kelapa sawit bergantung pada makanan dan sifat fizik habitat seperti yang ditentukan dengan

prinsip komponen analisis (PCA). Vermikompos dari PR dicampur dengan tandan kelapa sawit kosong (EFB) menggunakan *Eisenia fetida* di makmal memberikan pelarutan P yang tinggi dengan morocco PR (MPR) > gafsa PR (GPR) > togo PR (TPR) dari sistem kompos biasa. Nilai yang sama bagi pelepah kelapa sawit adalah 165, 52 dan 30%. Ini disebabkan oleh kandungan asid humik yang tinggi pada VPC (0.07g), populasi bakteria, Ca yang boleh diekstrak, dan enzim phosphomonoesterase pada kotoran cacing tanah. Bakteria dalam sistem pencernaan cacing-EFB adalah *Pseudomonas nitroreducens* dan *P. citronellolis*, dan C*ellulomonas flavigena* dalam cacing menggunakan pelepah kelapa sawit. Jumlah phenol yang dapat diekstrak sebanyak (10 g GAE 100 g^{-1} ekstrak) terdapat di awal proses pengkomposan EFB dan menurun setelah 4 minggu proses penguraian. Jenis sebatian yang terdapat dalam EFB segar adalah 2,4-bis(1,1-dimethyl)phenol, manakala yang telah di kompos adalah 2,6-bis (1,1-dimethyl)phenol. Sebaliknya tiada phenol didapati di dalam vermikompos EFB. Penggunaan terus 3 jenis cacing tanah di kajian rumah kaca menggunakan rumput *Setaria* tidak menunjukkan perbezaan yang bererti terhadap P tersedia, jumlah nutrient dalam tanah, pengambilan nutrient, dan hasil berat kering (DM) rumput. Penggunaan VPC menunjukkan pertumbuhan yang lebih baik bagi rumput *Setaria* berbanding EFB dan kompos konvensional. *Setaria* memerlukan sebanyak 75 tan ha^{-1} VPC untuk mencapai hasil berat kering maksimum. Penggunaan GPR dicampur dengan EFB, AM, dan cacing tanah meningkatkan berat kering rumput *Setaria* sebanyak 60% berbanding dengan GPR sahaja dan dengan menggunakan VPC (79%). Sumbangan AM, cacing tanah GPR terhadap pengambilan berbeza dan bergantung pada interaksi antaranya. Sebagai kesimpulannya, VPC adalah cara yang terbaik untuk menguruskan EFB. Kandungan nutriennya boleh menyokong pertumbuhan *Setaria*. Peningkatkan pertumbuhan rumput *Setaria* dan ketersediaan P dan nutrien lain (N dan K) ditunjukkan dengan kemampuan pengambilan nutrien yang diperolehi dengan mencampurkan EFB dengan cacing tanah, AM dan baja inorganik.

ACKNOWLEDGEMENTS

I would like to express my gratitude and appreciation to the following people for their contributions towards the completion of this thesis:

Prof. Dr. Mohamed Hanafi Musa, for his supervision, continual guidance, enthusiasm, encouragement, patience, and friendship during the course of my study.

Assoc.Prof. Dr.Mahmud Tengku Muda, for valuable discussion and constructive criticism.

Dr. Nor Azwady Abd Aziz, for his supervision, valuable suggestions, patience and friendship.

All staff in the Department of Land Management, for their assistance and friendship throughout the course of my study.

All Indonesian friends at UPM, for their friendship.

Finally, but most important, my family, for their prayers, love and continual support.

I certify that an Examination Committee has met on the April 4, 2007 to conduct the final examination of Tengku Sabrina Djunita on her Doctor of Philosophy thesis entitled "Enhancement of P Solubilization and Plant Nutrient Availability from Phosphate Rocks Through Vermicomposting and Arbuscular Mycorrhiza" in accordance with Universiti Pertanian Malaysia (Higher Degree)Act 1980 and Universiti Pertanian (Higher Degree) Regulations 1981. The Committee recommends that the candidate be awarded the relevant degree. Members of the Examination Committee are as follows:

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DECLARATION

I hereby declare that the thesis is based on my original work except for the quotations and the citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UPM or other institutions.

TENGKU SABRINA DJUNITA

Date: 3 May 2007

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LIST OF ABBREVIATIONS

AA	Auto-analyzer
AAS	Atomic absorption spectrophotometer
AM	Arbuscular mycorrhiza
BSTFA	N.O-bis(trimethyl-silyl)triflouroecetamide
DCM	Dichloromethane
DM	Dry matter
EFB	Empty fruit bunch
ESDR	Earthworm stocking density ratio
GAE	Gallic acid equivalent
GC-MS	Gas chromatography mass spectrometer
GPR	Gafsa phosphate rock
MPR	Morocco phosphate rock
PCA	Principle component analysis
PR	Phosphate rock
TPR	Togo phosphate rock
TSP	Triple super phosphate
UPM	University Putra Malaysia
VPC	Vermiphosphocompost

CHAPTER 1

INTRODUCTION

Phosphorus (P) is one of the key nutrients limiting plant growth. Its availability to plants is controlled mainly by soil pH, soluble Al, Fe, Ca and organic matter content. Most of soils in Malaysia are acidic, predominantly of Ultisols and Oxisols. These soils showed greater capability to fix phosphate, through their Fe and Al contents. Hence, added P fertilizer is strongly immobilized. Therefore, regular fertilizer applications are required to maintain adequate supply of plant-available P. In this situation the use of phosphate rock (PR) as a direct-application fertilizer could be the most cost effective than a water-soluble P source. Moreover, the escalation in costs of manufacturing and distribution of soluble P fertilizers made PR the cheapest and important alternative source for P. The solubility of PR in the soil is affected by several factors, such as the type of PR, soil factors (chemical, physical and biological), and climatic condition. The types of PR and soil factors could be modified, for example by adding lime into the soil to increase soil pH. Modification of pH could shifted P form from non soluble into soluble form. Several alternatives have been used to increase P availability from PRs including: (i) incorporation of additives into PR, (ii) partial acidulation of PR, (iii) compaction of PR with water-soluble P fertilizers, and (iv) microbial methods.

Many other materials including organic materials could be used to modify soils, and subsequently increase solubility of P in soil. The organic materials and their decomposition products can reduce P fixation in soils. These are attributed to the reduction in P fixation due to the complexation of Al and Fe by organic acids, competition between organic acids and orthophosphate for adsorption sites and release of P by organic material during decomposition.

As a major crop in Malaysia, oil palm produces by products in large quantities. Meanwhile, to maintain the production of fresh fruit bunch (FFB), oil palm trees need to be fertilized intensively using chemical fertilizers. By considering the large amount of these by-products they can be used to replace the nutrients lost by plant uptake. The total oil palm plantations area is around 4,051,374 ha in 2005 and expected to increase to 5.10 million ha in 2020 (Anonymous, 2005). For every tonne of crude palm oil (CPO) produced, three tonnes of palm oil mill effluent (POME) is generated. An average of 24 fronds is pruned every year, or equal to 11.7 tonnes ha^{-1} $year^{-1}$ (Chan, 2000). About 18.88 tonnes ha^{-1} y^{-1} of FFB was produced in 2005 and has generated 11.3 tonnes EFB ha^{-1} y^{-1} (Anonymous, 2005).

Thus, oil palm by-products are potential sources of nutrients. However, oil palm by-products are decomposed slowly in the environment. Soil micro and macro-organisms are required to enhance decomposition process. Soil macro-organisms, such as earthworm have a great potential to increase the soil productivity in oil palm plantation. Its role in P turnover is well known, about four to tenfold increase in P content and availability in earthworm casts result from enhanced mineralization of organic P (Sharpley and Syers, 1977). Inoculation of earthworms into the organic wastes during composting helps to enhance the transformation of organic P into mineral forms. In addition, they also keep the magnitude of fixation of released P into insoluble inorganic forms at low levels, thus increasing the availability of P in these compounds. Moreover, most of the additional P present in casts is held in more physically sorbed than chemisorbed forms and a reduction in P sorption capacity of soil by organic matter blockage. At the same time, the presence of earthworm will enlarge the microorganism's population in the soil.

Plant roots are the most important organ for nutrients and water absorption and uptake by plants. In the case of abnormal roots growth, the nutrients uptake will impede. Association of arbuscular mycorrhiza (AM) with roots would give plant with vigorous growth of root systems, hence plant will able to absorb more nutrients especially for immobile nutrient like P. Besides that, mycorrhizal infected plants are expected to dissolve P from PR.

In view of the role of earthworms and the amount of agriculture by products produce in oil palm plantation and the lack information about earthworm under oil palm tree ecosystems, this research was carried out in several oil palm plantations in Malaysia, to determine the suitability of using earthworm in decomposition of oil palm by-products, such EFB. The aims of this study were (i) to characterize earthworm populations and cast properties in different soil types and oil palm ages, and the relationships among soil properties on earthworm populations in the oil palm plantation, (ii) to evaluate the effect of vermicomposting on phosphate solubility of different P sources, (iii) to investigate the effect of application of EFB on earthworm population and activity under oil palm field condition, (iv) to evaluate the effect of vermiphosphocompost as a source of P fertilizer in fulfilling P requirement for plant growth compared to other sources, and (v) to evaluate the contribution of AM and earthworm in increasing P uptake by plant.

CHAPTER 2

LITERATURE REVIEW

2.1. Phosphate in Soil

Phosphate is a critical element in natural and agricultural ecosystems throughout the world. The concentration of P in the soils is in the range of 100-3,000 mg P kg^{-1} and the solubility is less than 0.01 mg PL^{-1} (Brady and Weil, 2000) or around 0.1% of the total P. Most of soils have low P availability and in order to fulfill the plants P requirement, large quantities of P fertilizer and P-containing organic wastes have frequently been added.

The main sources of P in soil are from inorganic and organic phosphates. About 150 minerals are known to contain at least 0.44% P (1% P_2O_5). The world's supply of P comes from mineral deposits. The important inorganic phosphate source minerals is apatite group $Ca_{10}(PO_4, CO_3)_6(F,OH)_{2-3}$ and it is the minable deposits. If this apatite-bearing rock is high enough in P content to be used directly to make fertilizer or as a furnace charge to make elemental P, it is called phosphate rock (Cathcart, 1980).

Based on their geological formation, the present phosphate rock mined can be divided into sedimentary, igneous and metamorphic rocks origin (McCleellan and Gremillion, 1980; Slansky, 1986).